Stephen G. Withers *et al*. Application No.: 09/837,711

Page 2

## **AMENDMENTS TO THE CLAIMS:**

This listing of claims will replace all prior versions, and listings of claims in the application:

## **IN THE CLAIMS:**

## Claims 1-39 (Cancelled)

- 40. (Currently amended) A method for synthesizing an oligosaccharide comprising the steps of:
- (a) combining a <u>modified</u> glycosyl donor molecule and a glycoside acceptor molecule in a reaction mixture; and
- (b) enzymatically coupling the donor molecule to the acceptor molecule using a mutant form of glycosidase enzyme to form the oligosaccharide, said enzyme being selected from among glycosidase enzymes having two catalytically active amino acids with carboxylic acid side chains within the active site of the wild-type enzyme, and said mutant enzyme being mutated to replace one of said <u>catalytically active</u> amino acids having a carboxylic acid side chain with a different amino acid of comparable or smaller size, said different amino acid having a non-carboxylic acid side chain characterized in that, said <u>modified</u> glycosyl donor molecule having a  $\beta$  configuration and said glycoside acceptor molecule having an  $\alpha$  configuration, or vice versa.
- 41. (Currently amended) The method of claim 40, wherein the glycosidase enzyme is a stereochemistry retaining enzyme in which one of the carboxylic acid side chains chain of one of said catalytically active amino acids in the glycosidase enzyme active site functions as an acid/base catalyst and the other carboxylic acid side chain of the other

**PATENT** 

Stephen G. Withers *et al.* Application No.: 09/837,711

Page 3

<u>catalytically active amino acid</u> functions as a nucleophile, and wherein the amino acid having the nucleophile carboxylic acid side chain is replaced in the mutant enzyme.

- 42. (Previously presented) The method of claim 41, wherein the enzyme is a  $\beta$ -glycosidase.
- 43. (Previously presented) The method of claim 42, wherein the glycosyl donor molecule is an  $\alpha$ -glycosyl fluoride.
- 44. (Previously presented) The method of claim 43, wherein the  $\alpha$ -glycosyl fluoride is an  $\alpha$ -glucosyl fluoride.
- 45. (Previously presented) The method of claim 43, wherein the  $\alpha$ -glycosyl fluoride is a  $\alpha$ -galactosyl fluoride.
- 46. (Previously presented) The method of claim 40, wherein the enzyme is a  $\beta$ -glycosidase.
- 47. (Previously presented) The method of claim 40, wherein the enzyme is a  $\beta$ -glucosidase.
- 48. (Previously presented) The method of claim 40, wherein the acceptor molecule is an aryl-glycoside.
- 49. (Previously presented) The method of claim 48, wherein the acceptor molecule is a nitrophenyl-glycoside.
- 50. (Currently amended) The method of claim 40, wherein the glycosidase enzyme is a stereochemistry inverting enzyme in which one of the carboxylic acid side chains of one of said catalytically active amino acids in the active site of the glycosidase enzyme functions as an acid catalyst and the other carboxylic acid side chain of the other catalytically active amino acid functions as a base catalyst, and wherein the amino acid having the carboxylic acid die side chain which functions as a base catalyst is replaced in the mutant enzyme.

Stephen G. Withers *et al*. Application No.: 09/837,711

Page 4

## 51 - 54 (Cancelled)

55. (Previously presented) The method of claim 40, wherein the glycosidase enzyme is selected from the group consisting of  $\beta$ -glucosidases,  $\beta$ -galactosidases,  $\beta$ -mannosidases,  $\beta$ -N-acetyl glucosaminidases,  $\beta$ -N acetyl galactosaminidases,  $\beta$ -xylosidases,  $\beta$ -fucosidsases, cellulases, xylanases, galactanases, mannanases, hemicellulases, amylases, glucoamylases,  $\alpha$ -galactosidases,  $\alpha$ -mannosidases,  $\alpha$ -N-acetyl glucosaminidases,  $\alpha$ -N acetyl galactosaminidases,  $\alpha$ -xylosidases,  $\alpha$ -fucosidsases, and neuraminidases/sialidases.

56 - 70 (Cancelled)